

**STUDIES ON THE INTERACTIONS OF TOBACCO  
CONSTITUENTS WITH MONOAMINE OXIDASE A AND B**

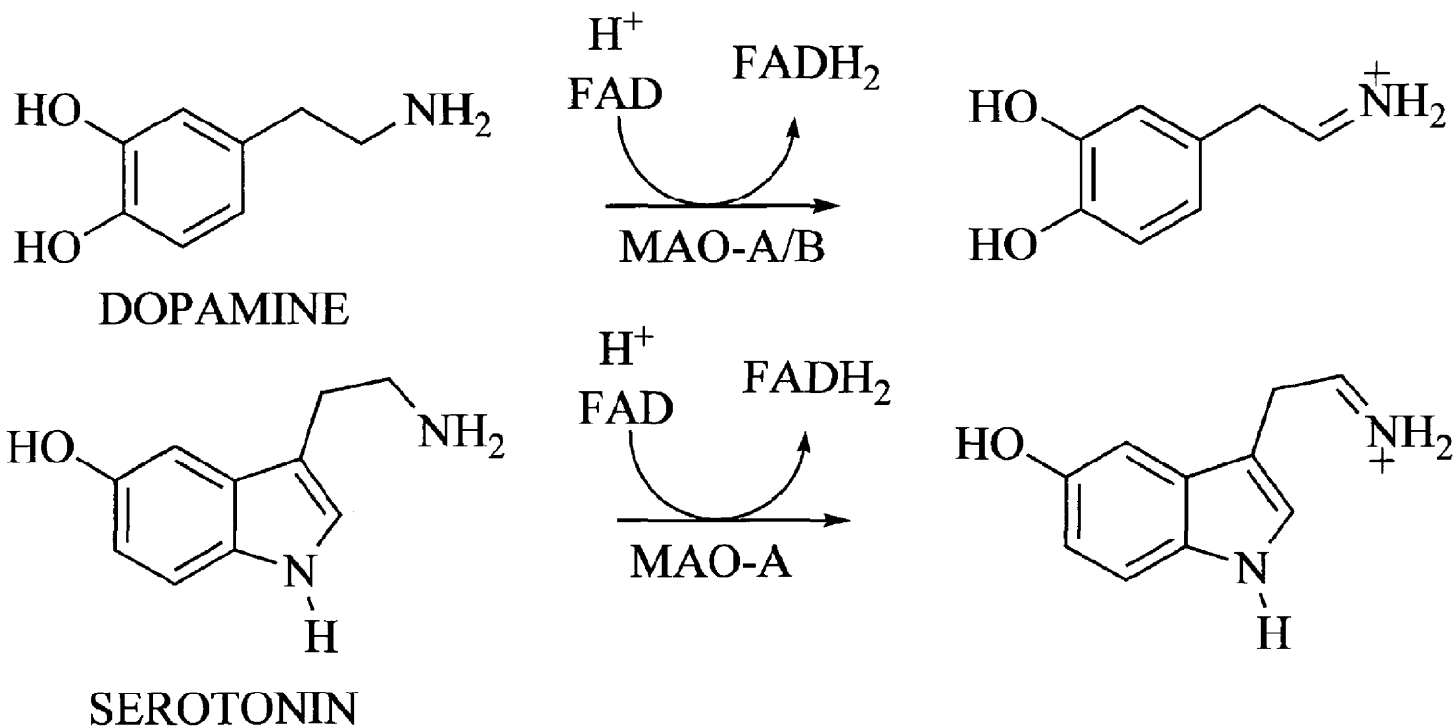
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Department of Chemistry  
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Blacksburg, VA

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## MONOAMINE OXIDASE CATALYZED OXIDATION OF DOPAMINE AND SEROTONIN



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**EFFECTS OF TOBACCO SMOKE EXPOSURE IN HUMANS ON  
MAO-A AND MAO-B AS REVEALED BY PET BRAIN SCANS**

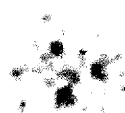
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# Imaging MAO B in the Human Brain



non-smoker (female 48 years old)

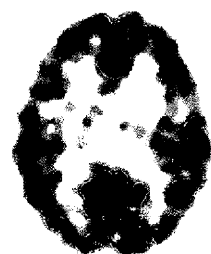


smoker (female 51 years old)

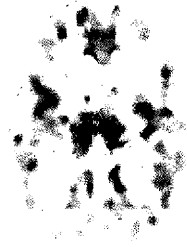
MAO B level ↑

L-deprenyl treatment (male, 43 years old)

# Brain MAO A and Smoking Status



Non-Smoker (male, 34 years)



Smoker (male, 31 years)



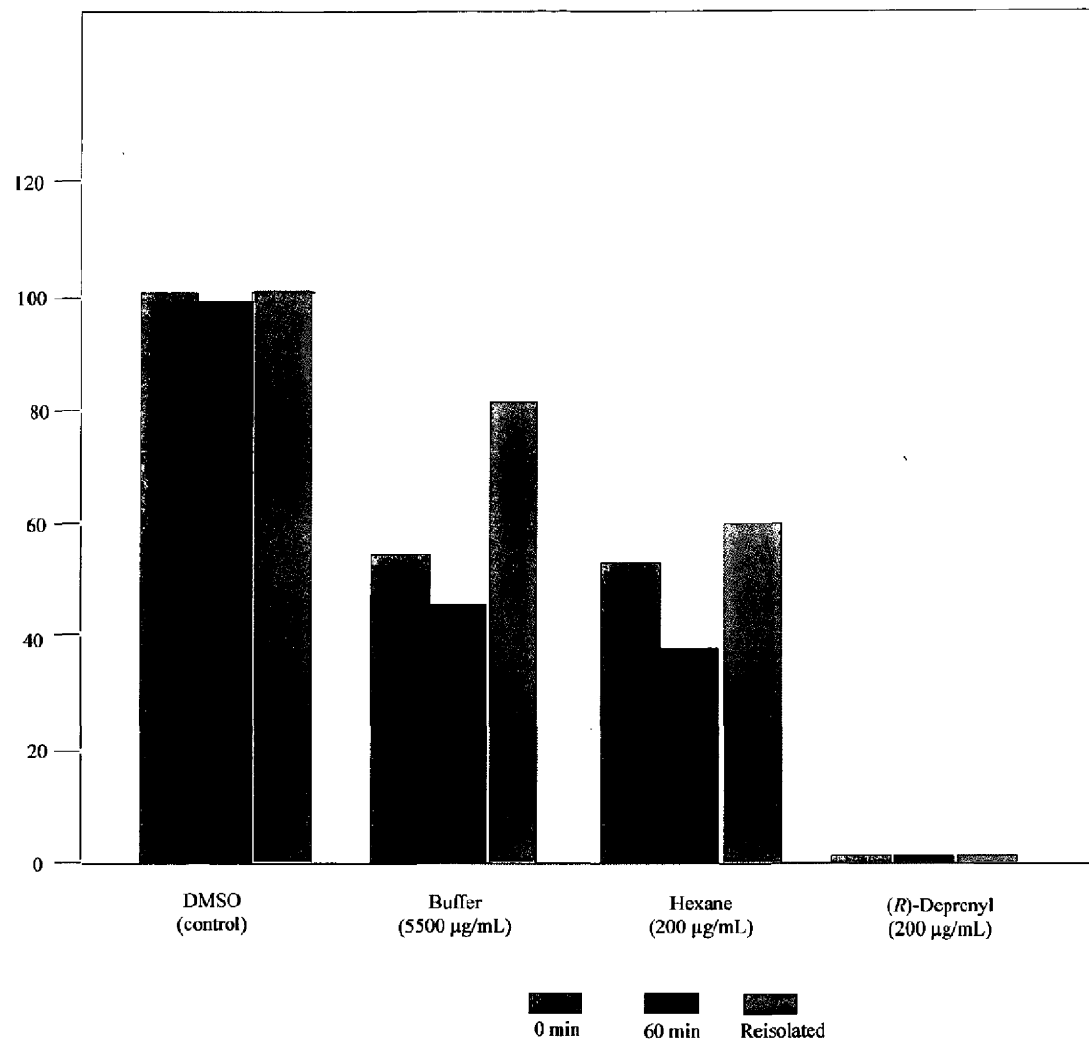
Tranylcypromine Treatment (male, 34 years)

*IN VITRO* ASSAY FOR MAO-B INHIBITOR  
ACTIVITY OF TOBACCO SMOKE EXTRACTS

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## INHIBITION OF MAO-B BY EXTRACTS FROM CIGARETTE SMOKE



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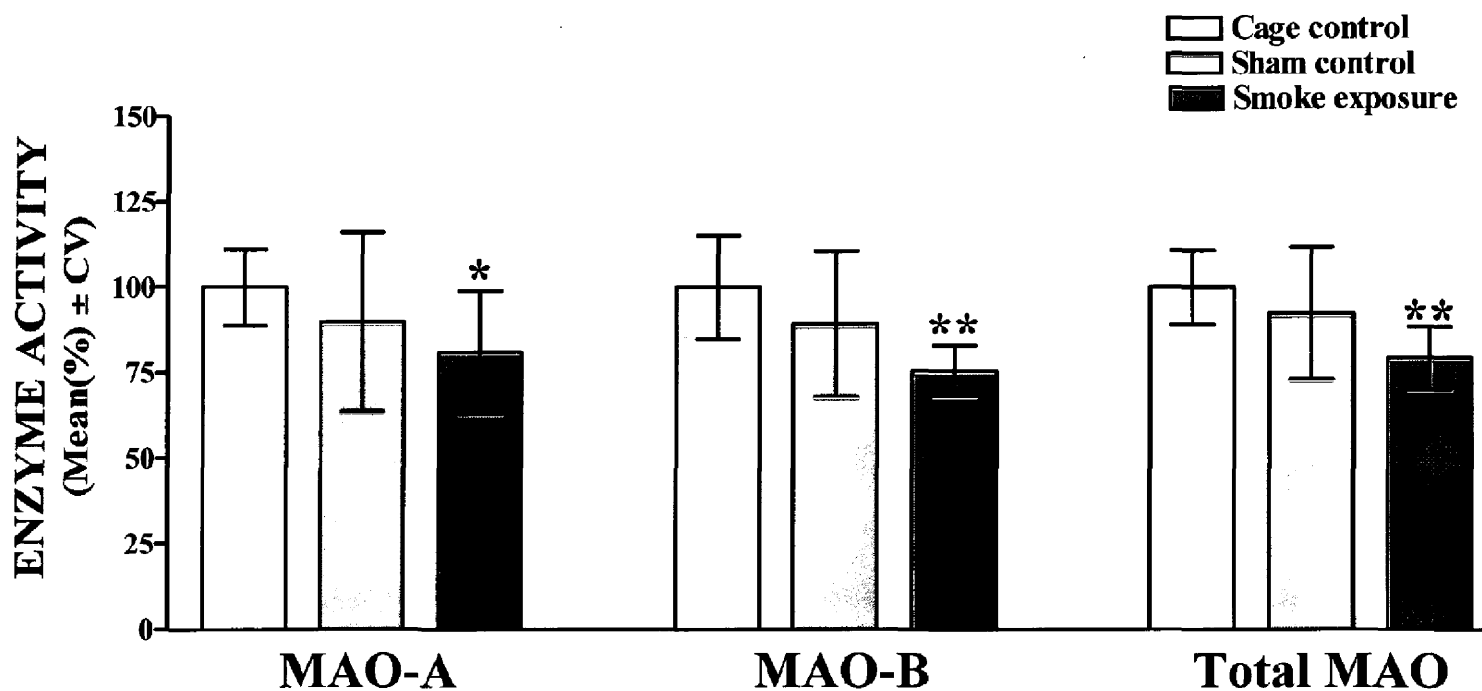
**EFFECTS OF SMOKE EXPOSURE ON SPRAGUE-  
DAWLEY RAT BRAIN MAO-A AND MAO-B ACTIVITIES**

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# Rat Brain MAO Activity: 12-Week Smoke Exposure vs. Control (n = 6)



\*p<0.05

\*\*p<0.005

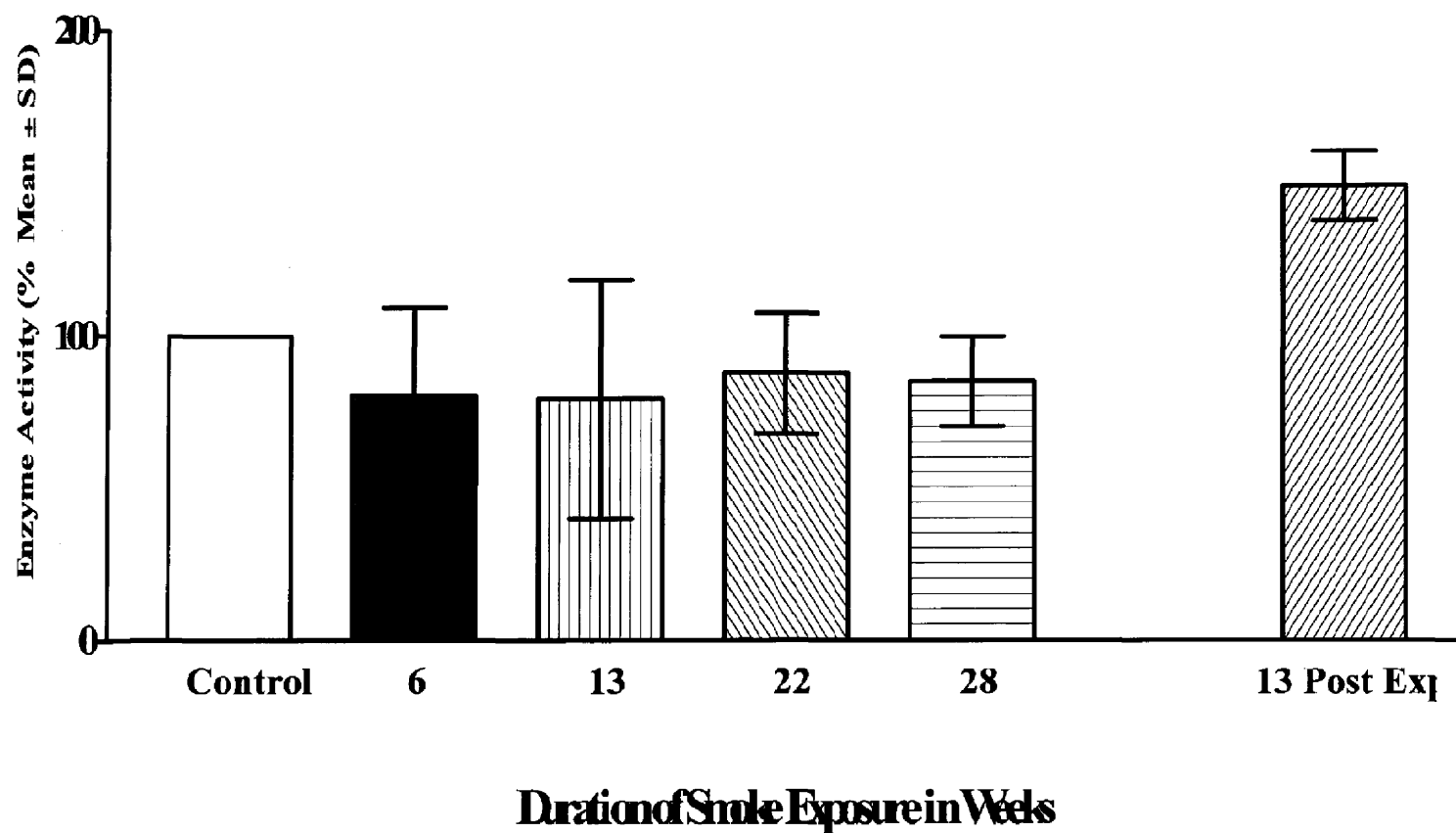
**EFFECTS OF TOBACCO SMOKE EXPOSURE  
ON BRAIN MAO-A AND MAO-B  
ACTIVITIES IN C57BL/6 MICE**

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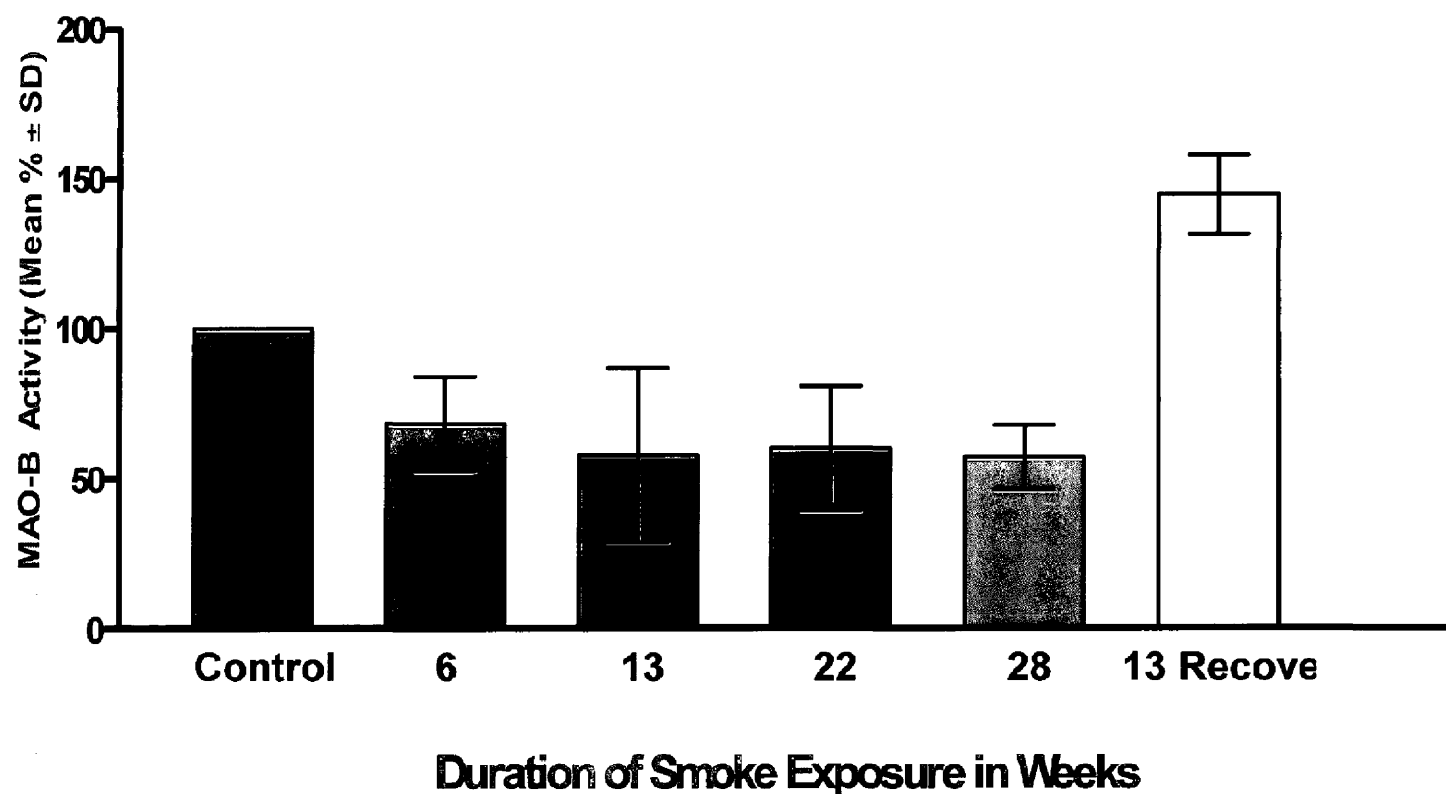
## MAO-A Activity of C57 Mouse Brain Mitochondrial Homogenates After Exposure to 600 $\mu\text{g/L}$ Smoke Vs Control



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## MAO-B Activity of C57 Mouse Brain Mitochondrial Homogenates After Exposure to 600 $\mu\text{g/L}$ Smoke Vs Control



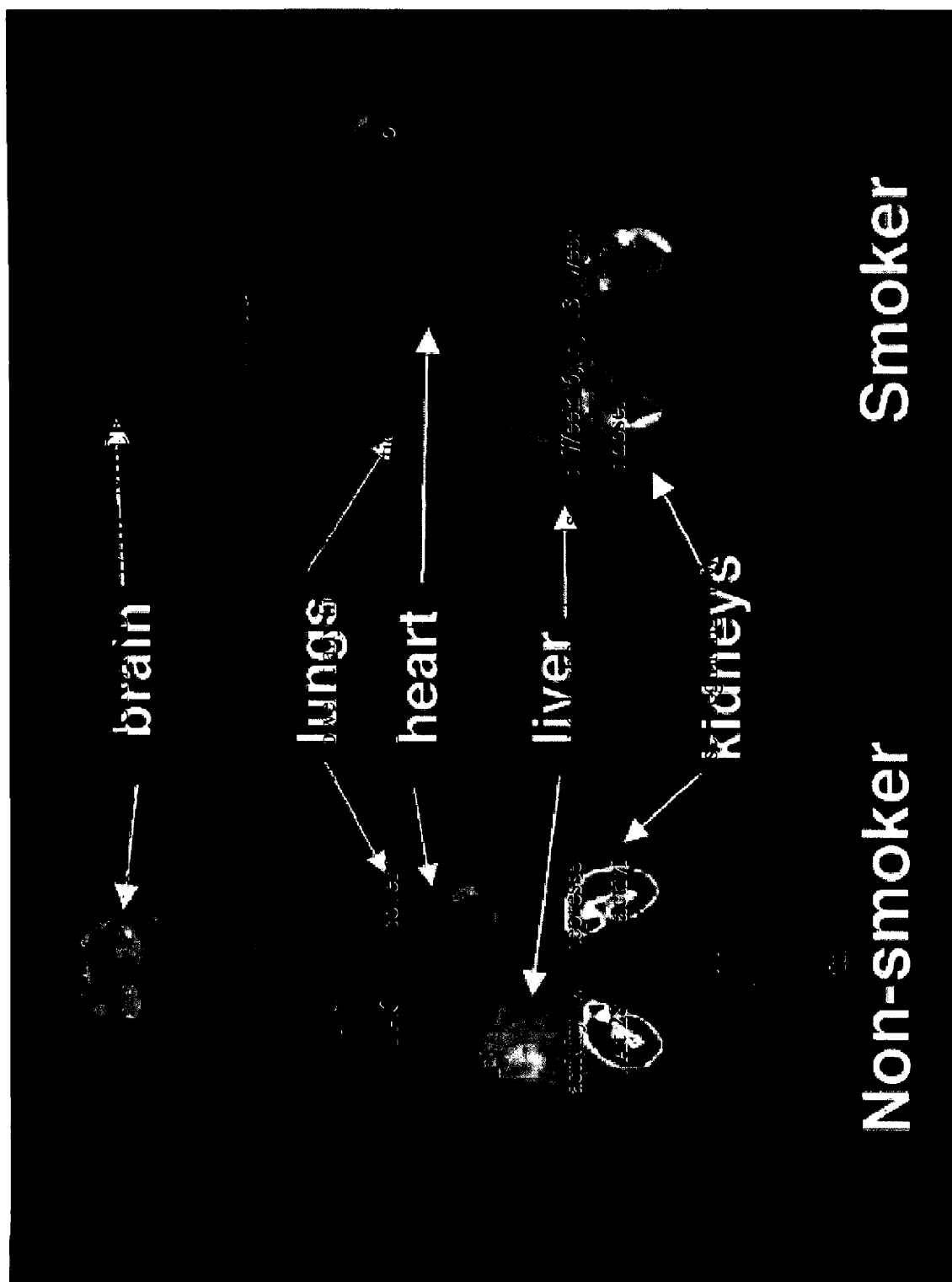
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**EFFECTS OF TOBACCO SMOKE EXPOSURE  
IN HUMANS ON MAO-B AS REVEALED  
BY WHOLE BODY PET BRAIN SCANS**

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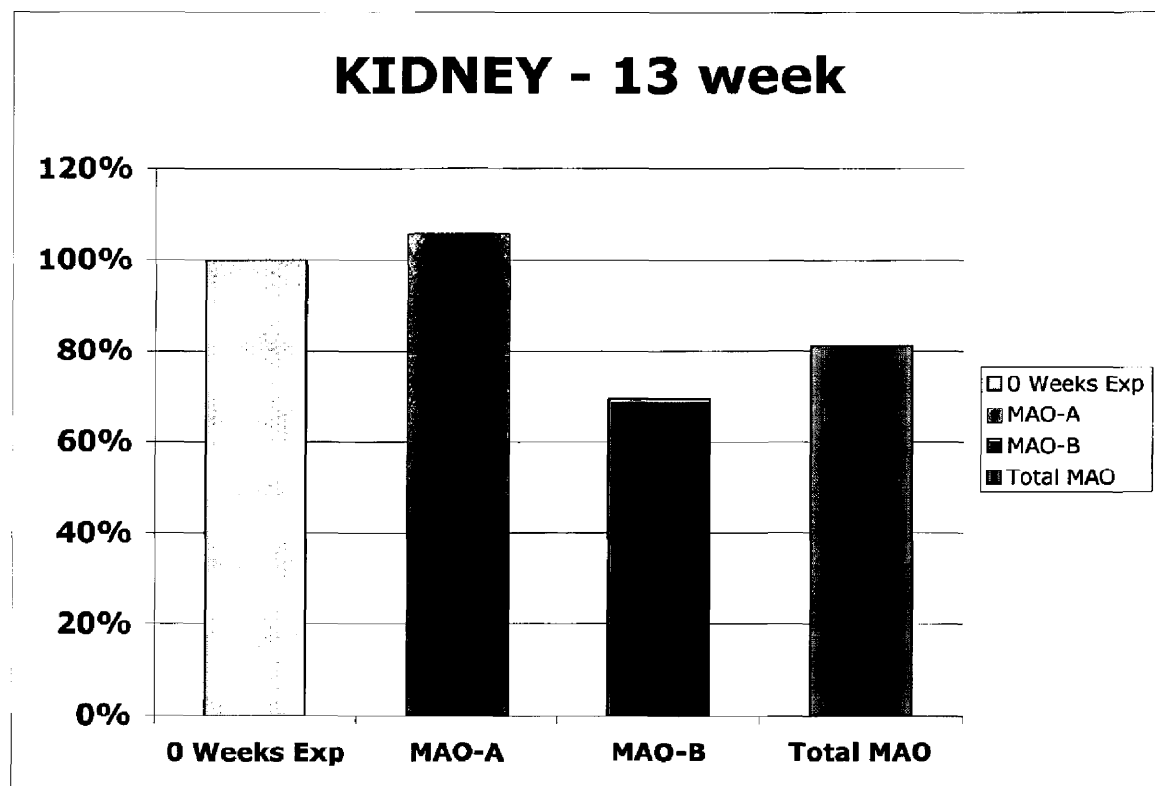
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**EFFECTS OF TOBACCO SMOKE EXPOSURE ON MAO-B  
ACTIVITY ON PERIPHERAL ORGANS:  
A 13-WEEK STUDY IN C57BL/6 MICE**

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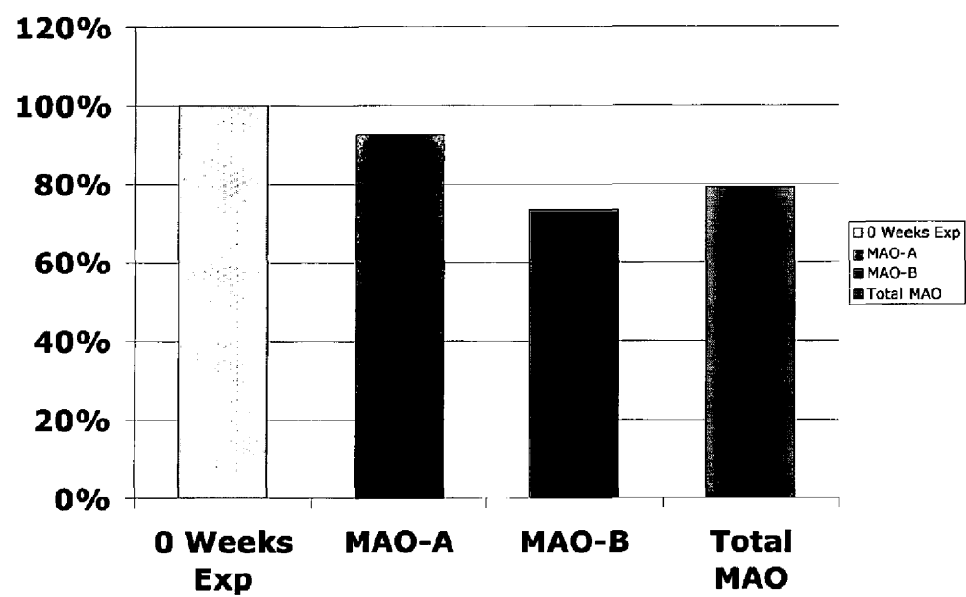


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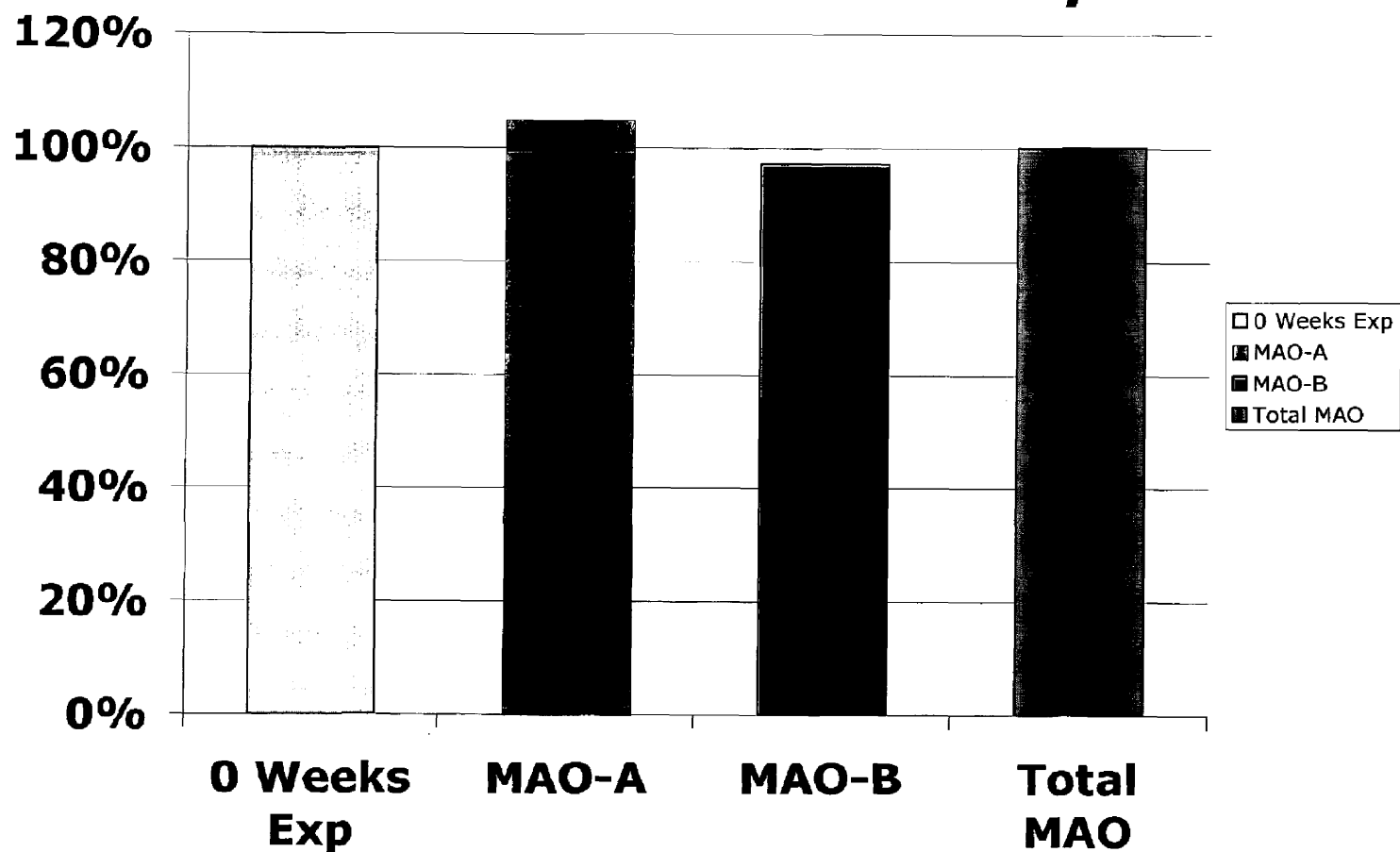
### KIDNEY - 1 Week Recovery



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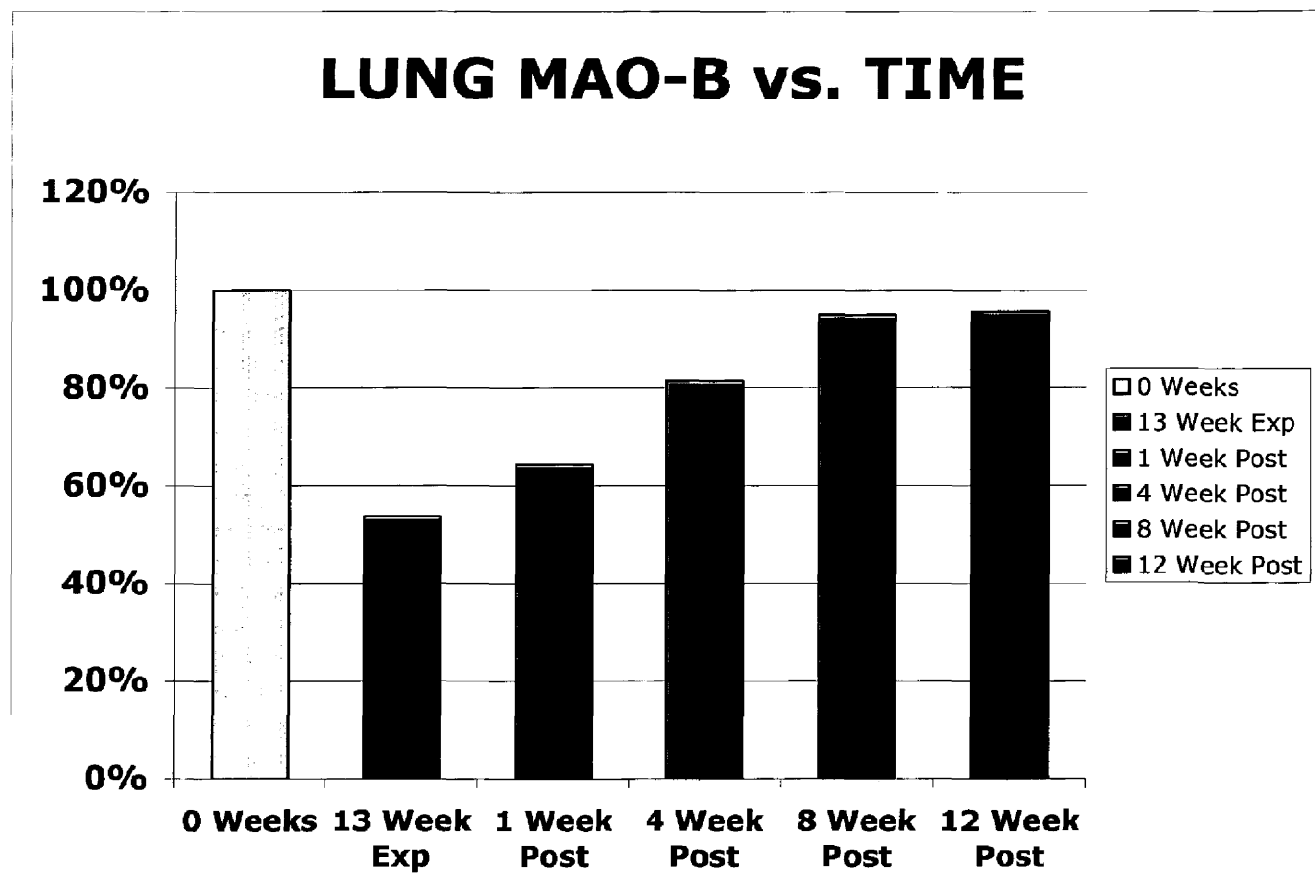
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## KIDNEY - 4 Week Recovery



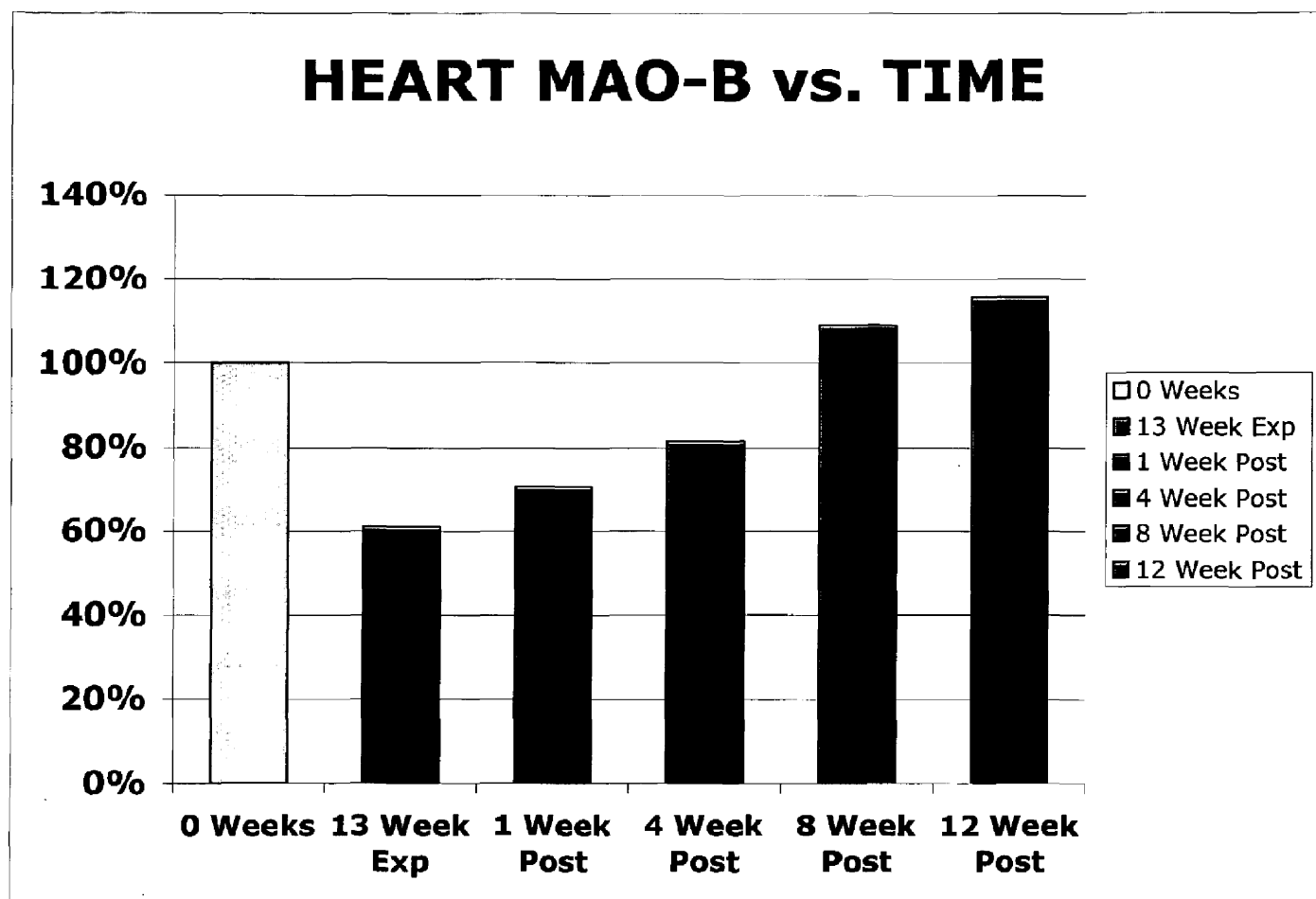
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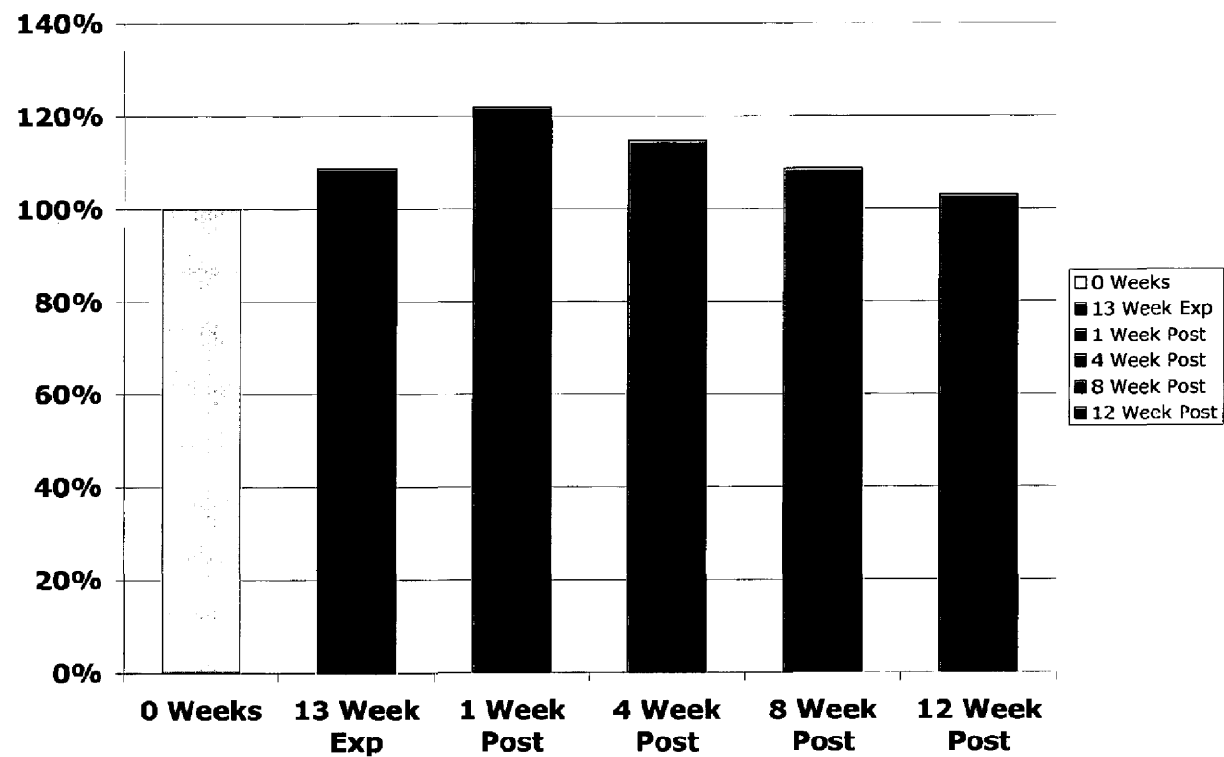
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## LIVER MAO-B vs. TIME



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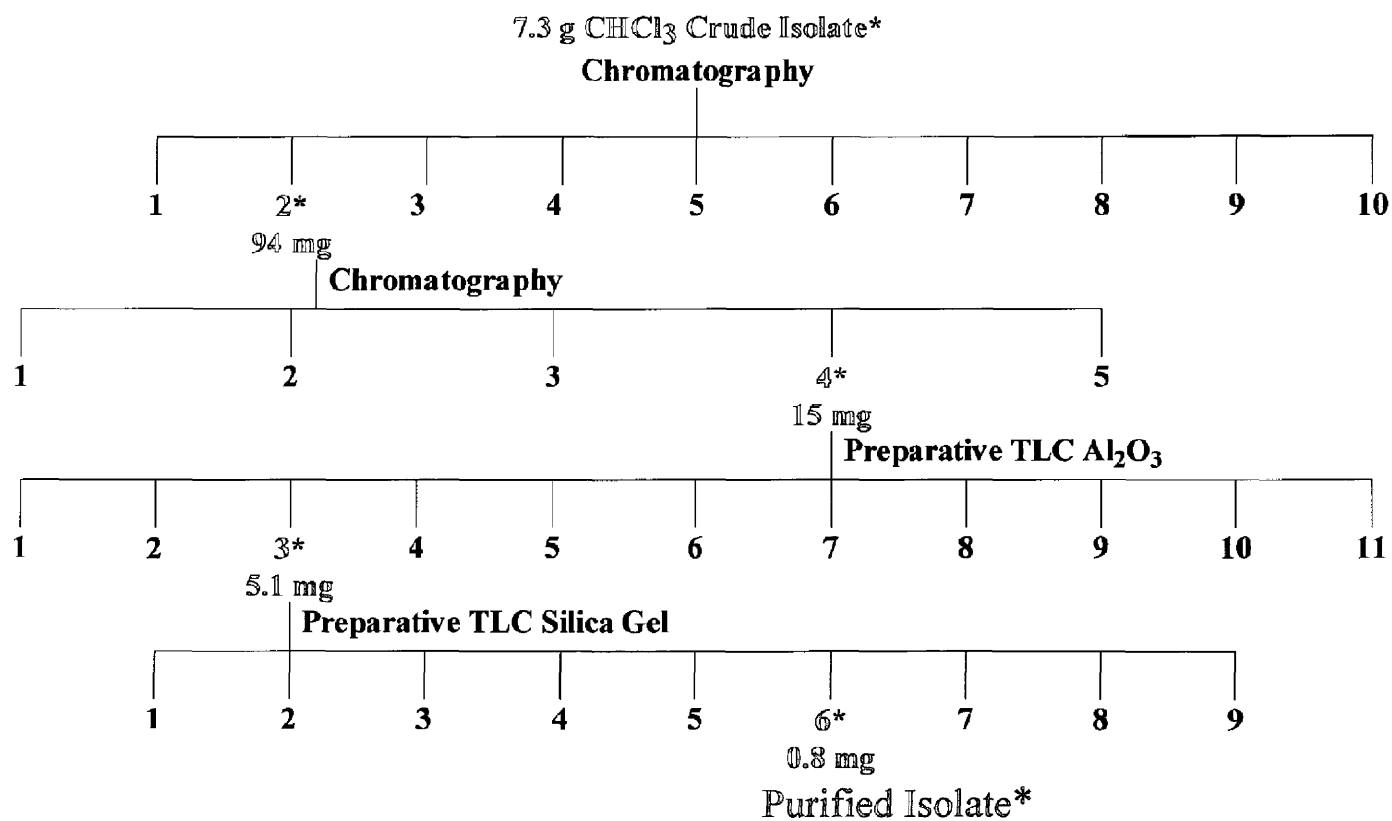
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# FRACTIONATION STUDIES

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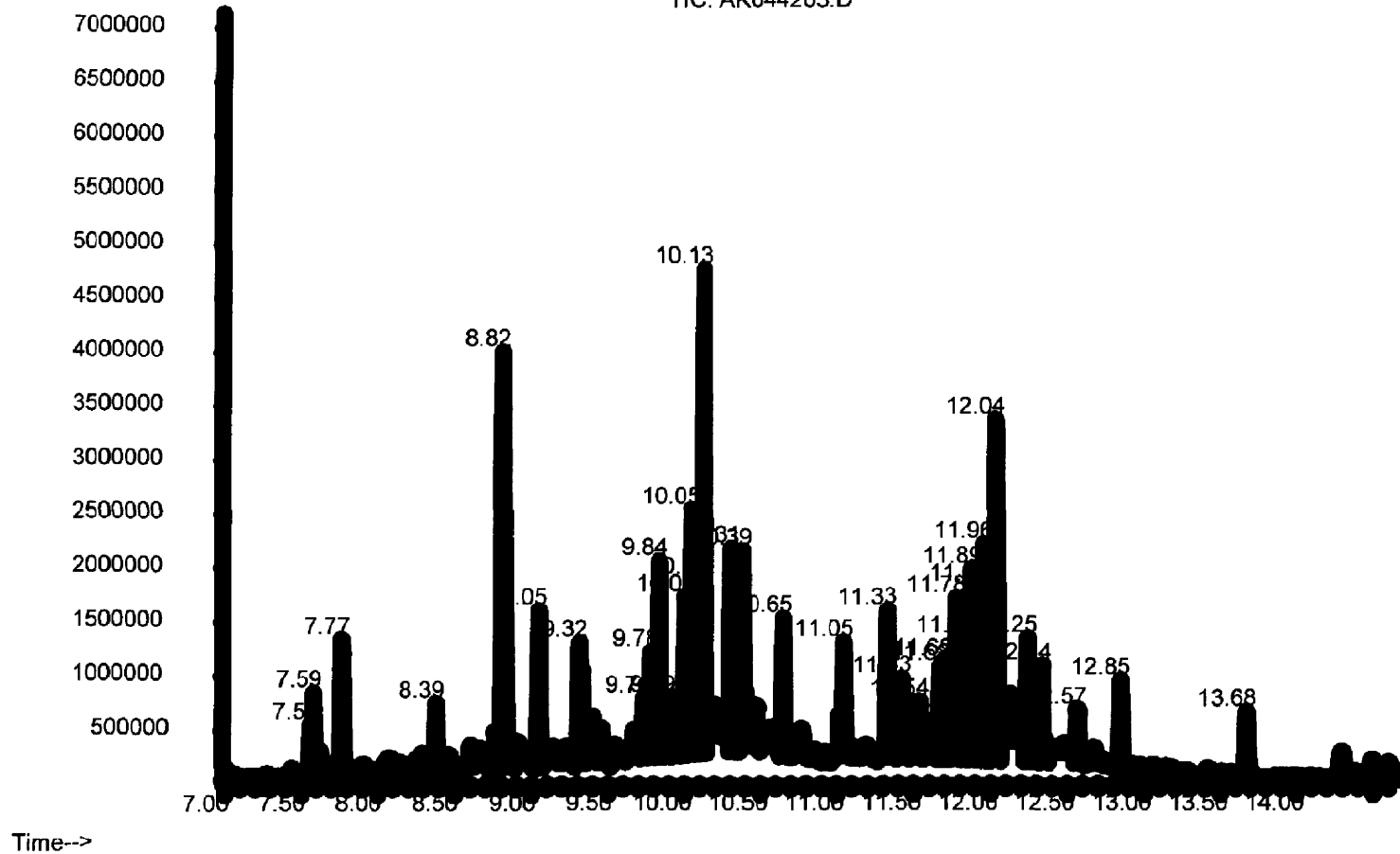
**BIOASSAY-GUIDED FRACTIONATION**

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Abundance

TIC: AK044203.D

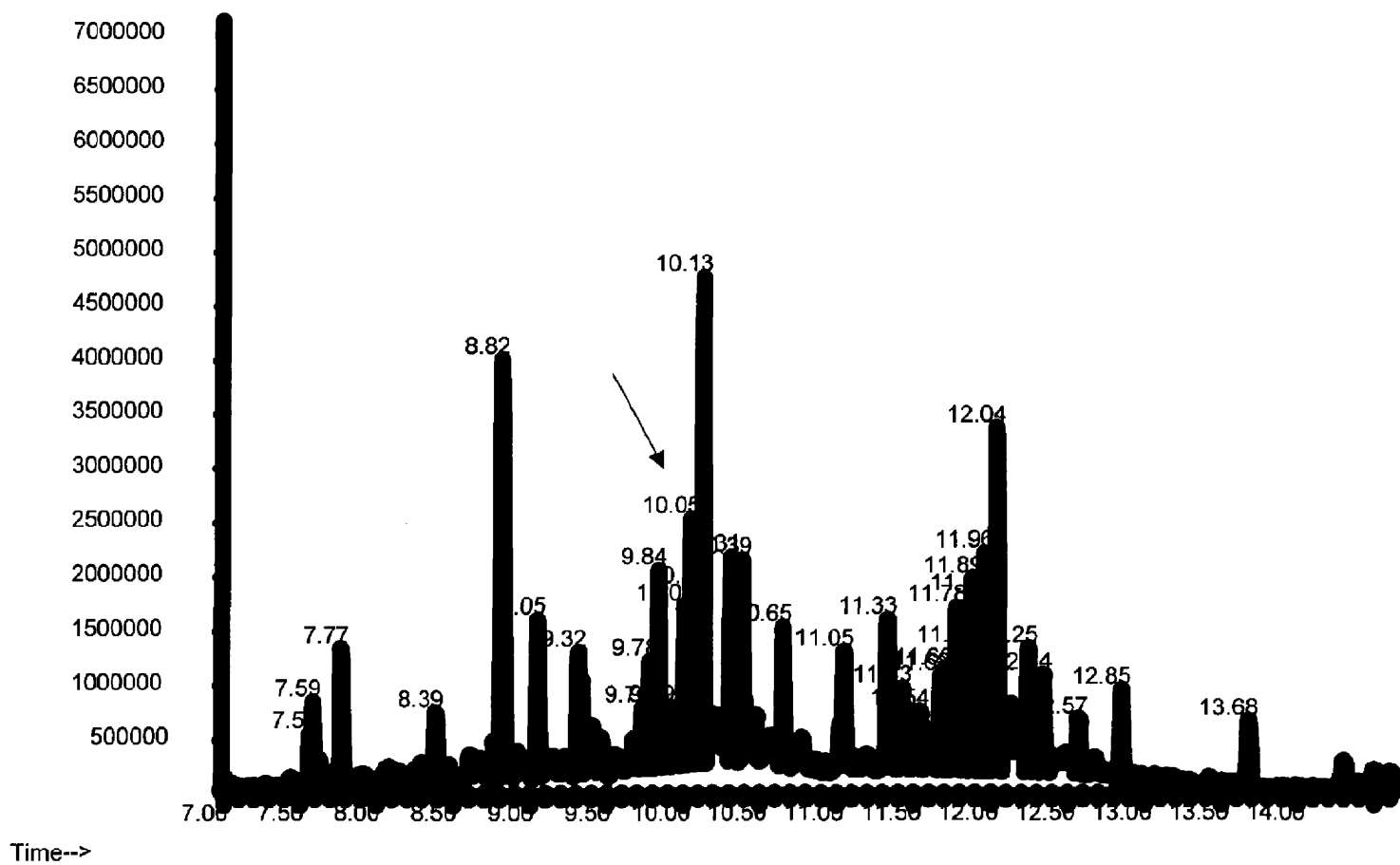


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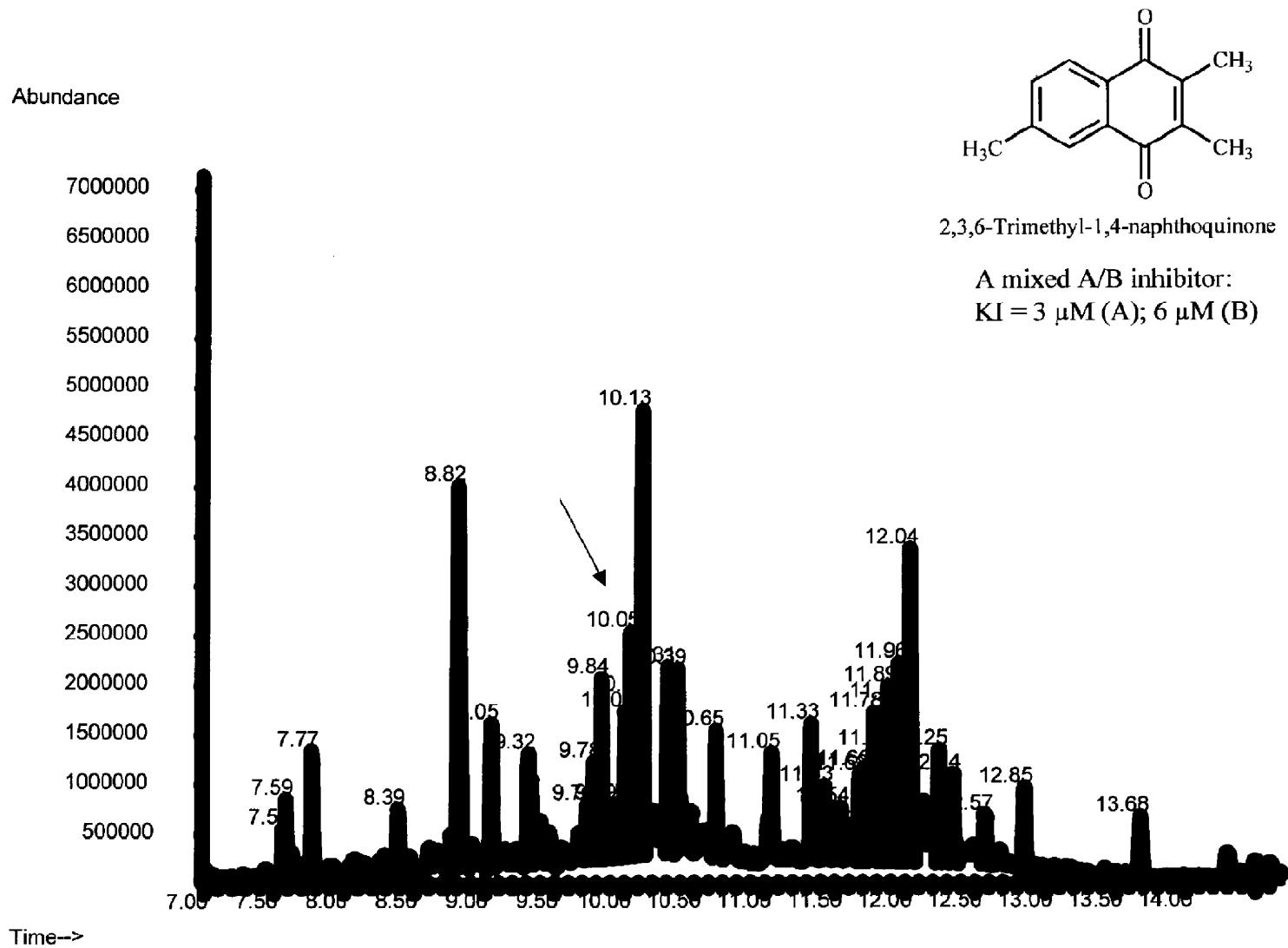


Abundance



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# The Combined Fractions

Neutral Alumina Column Chromatography (10 g)  
Evaluated at 10.0  $\mu\text{g/mL}$  (KHA-3-110)

Fraction 1	Fraction 2	Fraction 3	Fraction 4	Fraction 5	Fraction 6	Fraction 7	Fraction 8
5.44 mg	4.70 mg	9.51 mg	14.35 mg	25.58 mg	7.38 mg	3.63 mg	3.01 mg
66.9 %	63.7 %	41.6 %	62.6 %	18.7 %	21.5 %	32.8 %	2.8 %

Neutral Alumina Column Chromatography, 0.8 g  
Evaluated at 6  $\mu\text{g/mL}$  (KHA-3-120)

Fraction 1	Fraction 2	Fraction 3	Fraction 4	Fraction 5
1.76 mg	1.89 mg	1.97 mg	1.50 mg	1.16 mg
57.0 %	72.1 %	74.7 %	43.0 %	23.8 %

SPE-Column (C-18)  
Evaluated at 10  $\mu\text{g/mL}$

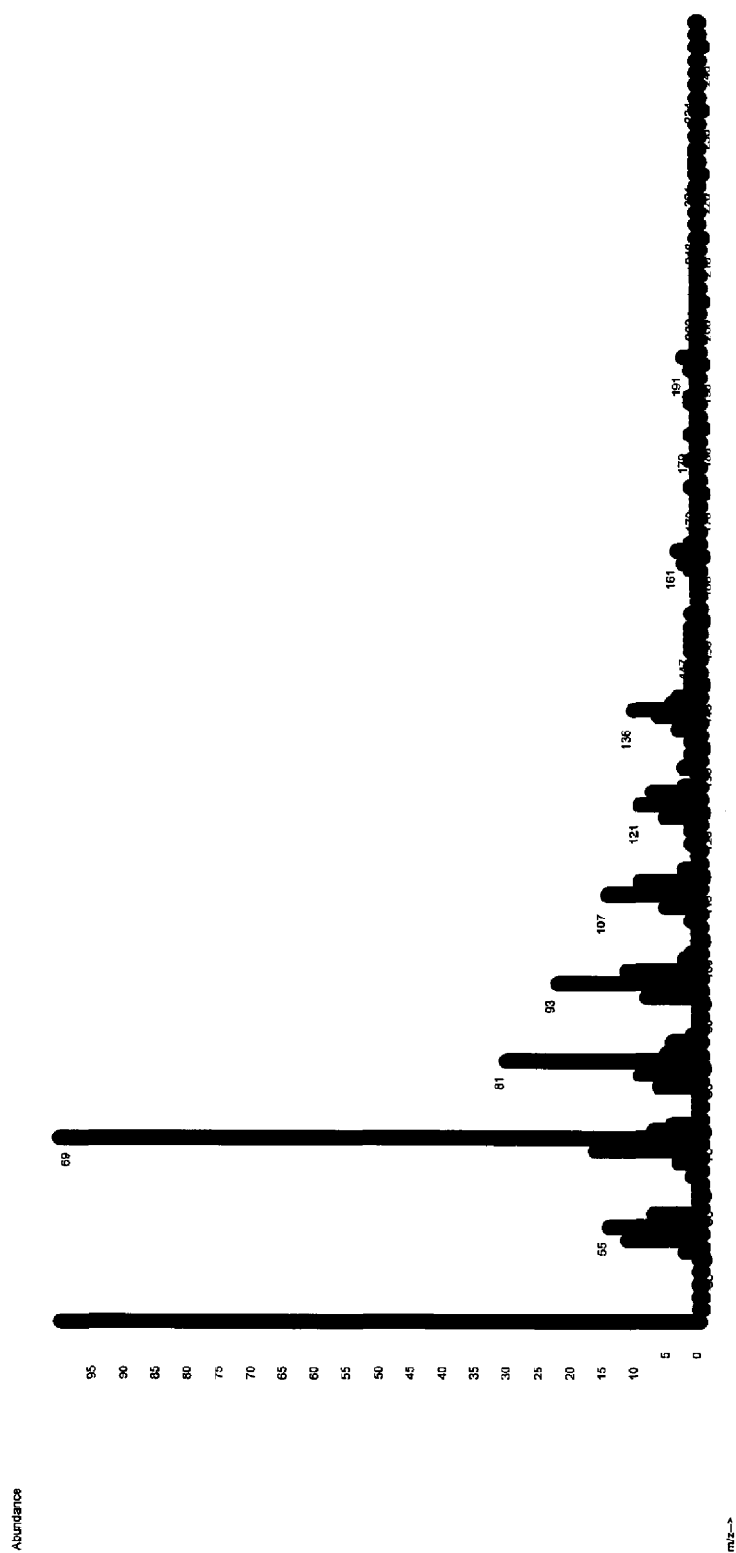
1	2	3	4	5
----	3.93 mg	2.29 mg	0.27 mg	0.56 mg
	50.8 %	49.6 %	32.3 %	34.4 %

SPE-Column (C-18)  
Evaluated at 10  $\mu\text{g/mL}$

1	2	3	4
0.68 mg	1.1 mg	0.31 mg	0.55 mg
36 %	61.7 %	86.8 %	14.8 %

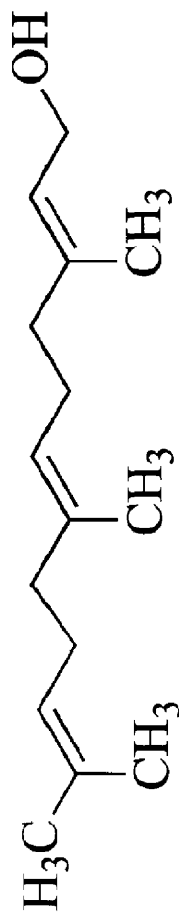
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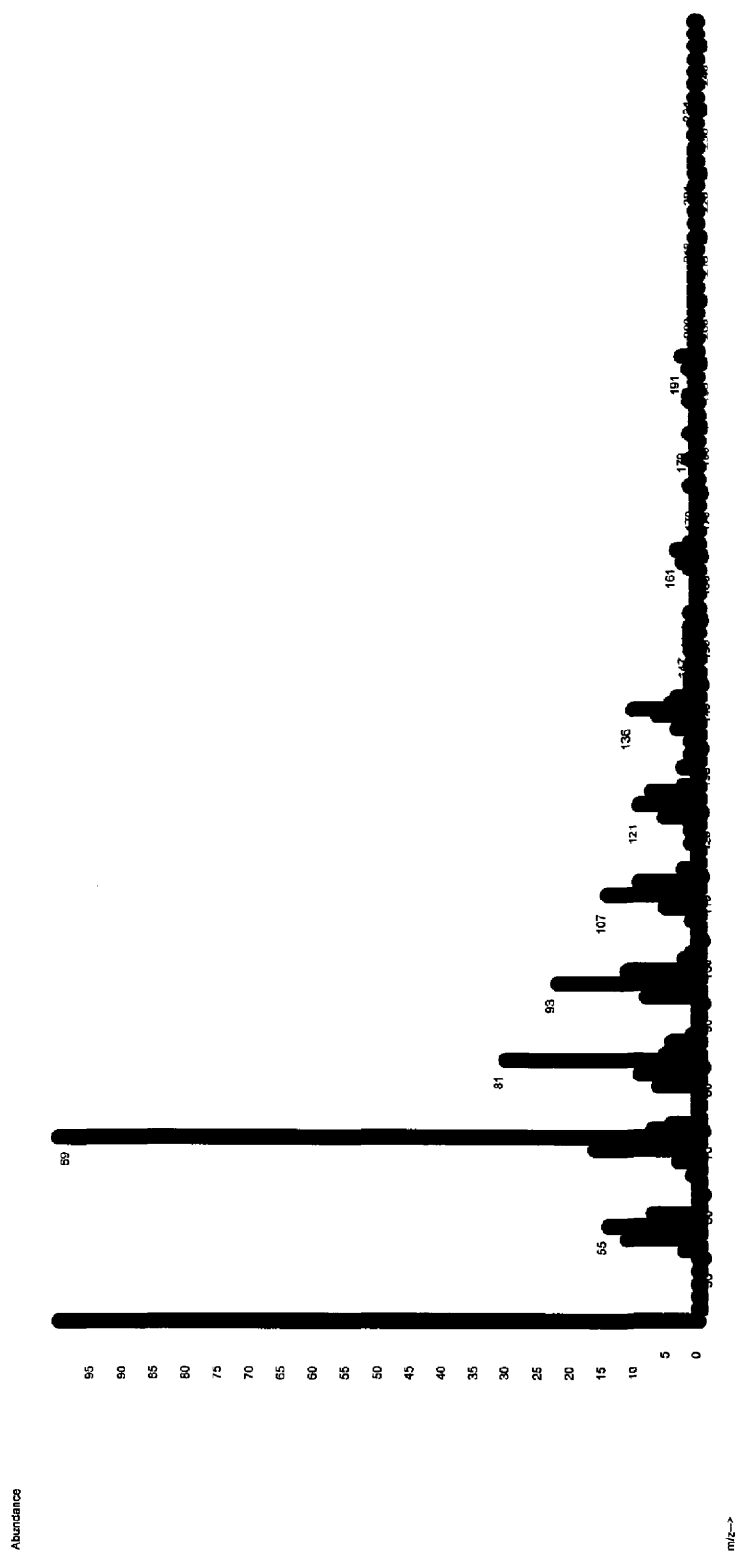


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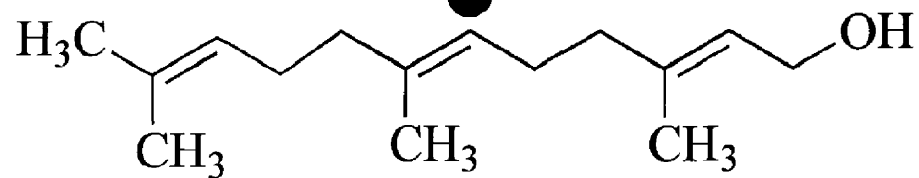


**Farnesol**



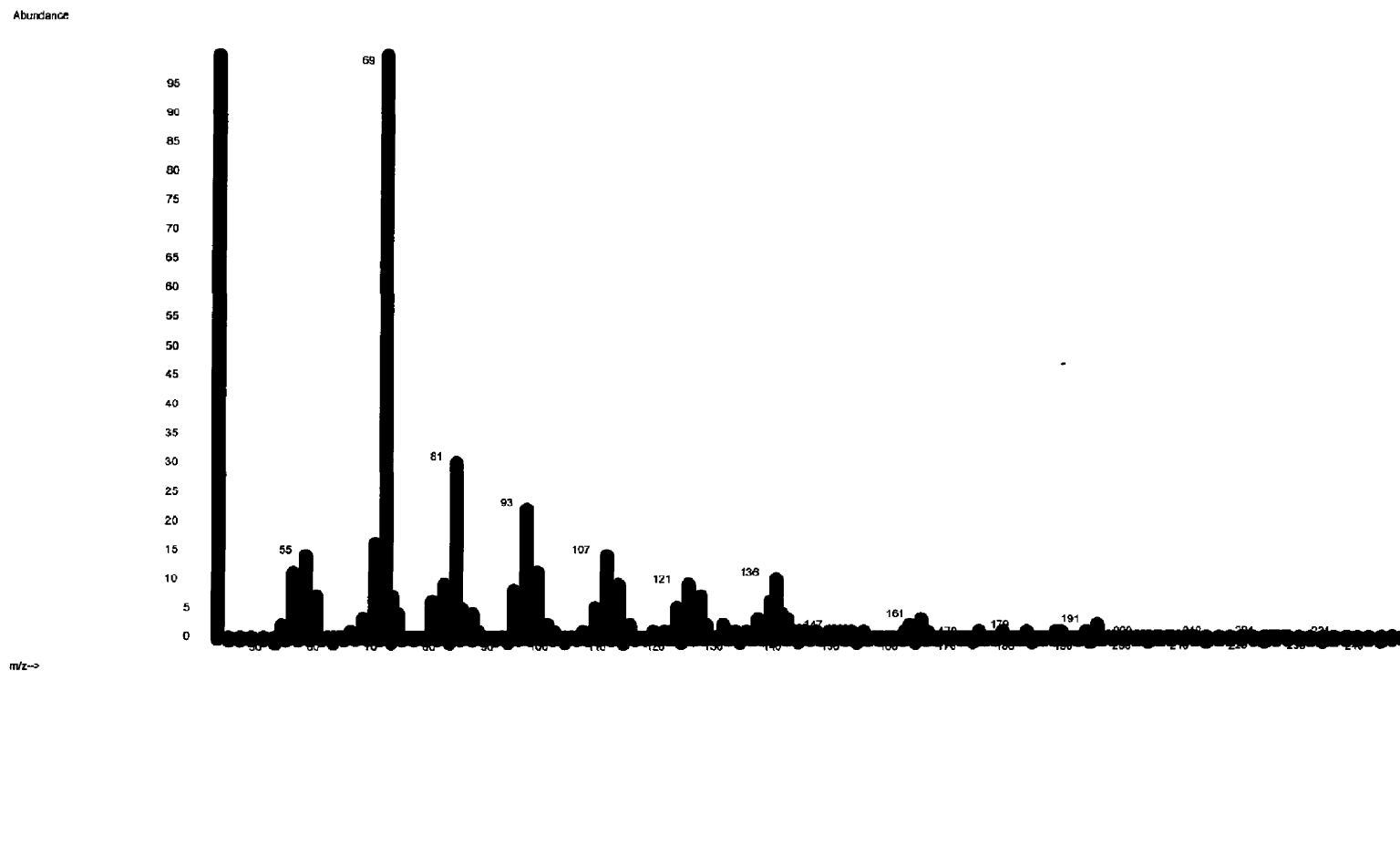
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## Farnesol

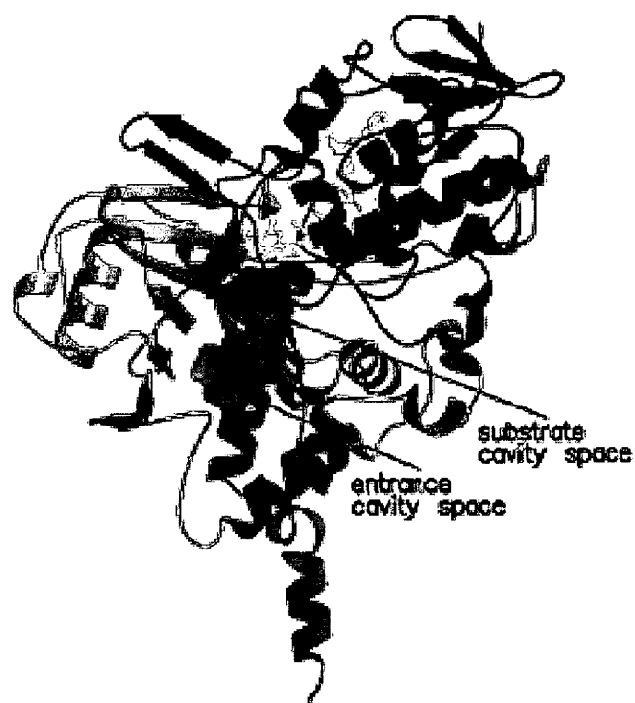
A HUMAN MAO-B SELECTIVE INHIBITOR ( $K_I < 1 \mu\text{M}$ )



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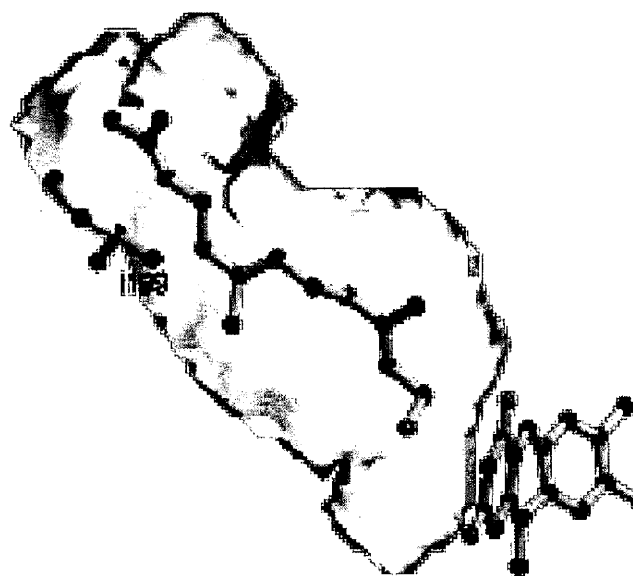
X-RAY STRUCTURE OF *TRANS,TRANS*-FARNESOL  
OCCUPYING THE ACTIVE SITE OF HUMAN MAO-B



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JUSTAPOSITION OF THE OH GROUP OF FARNESOL  
TO THE FLAVIN COFACTOR OF HUMAN MAO-B



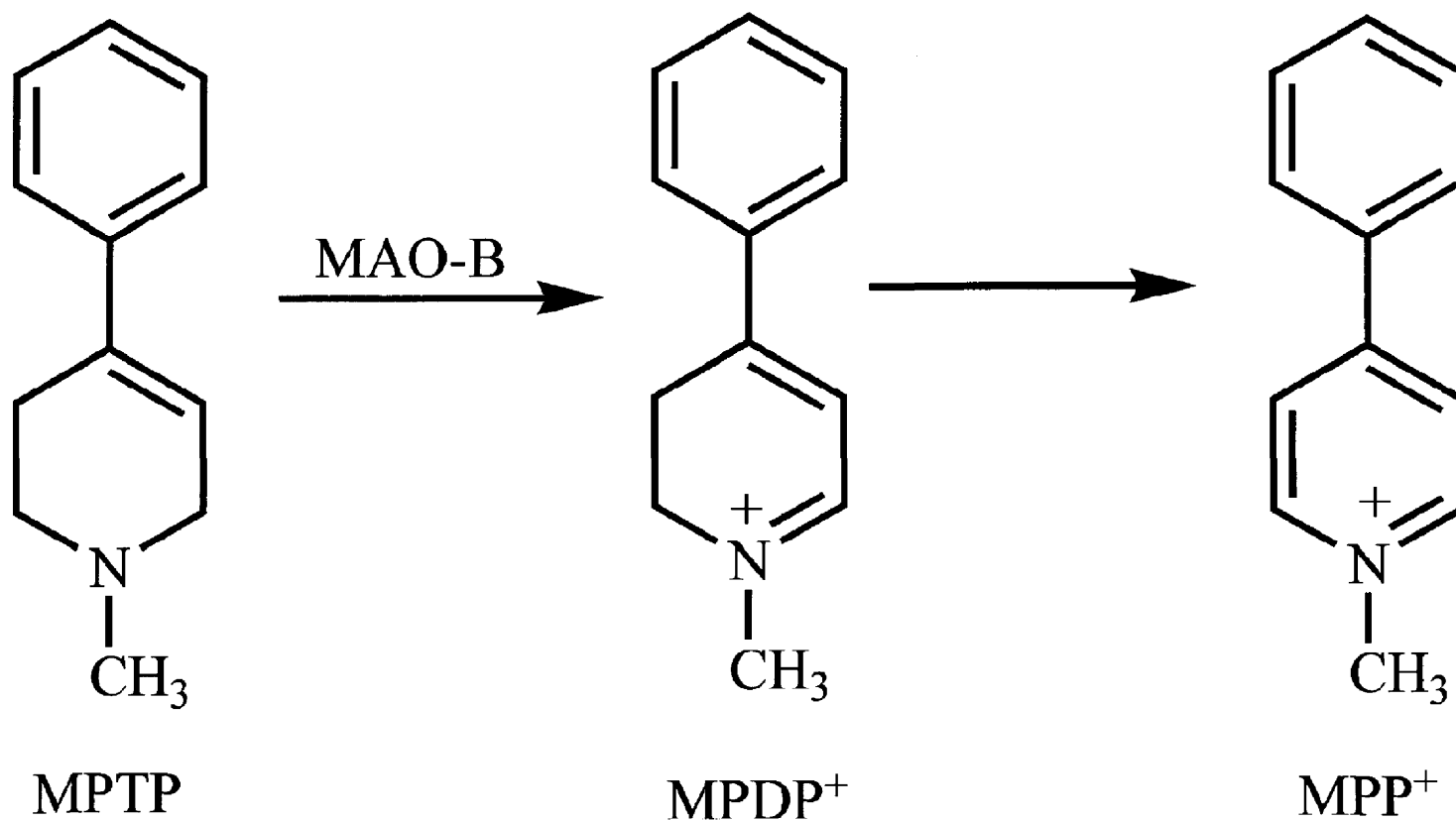
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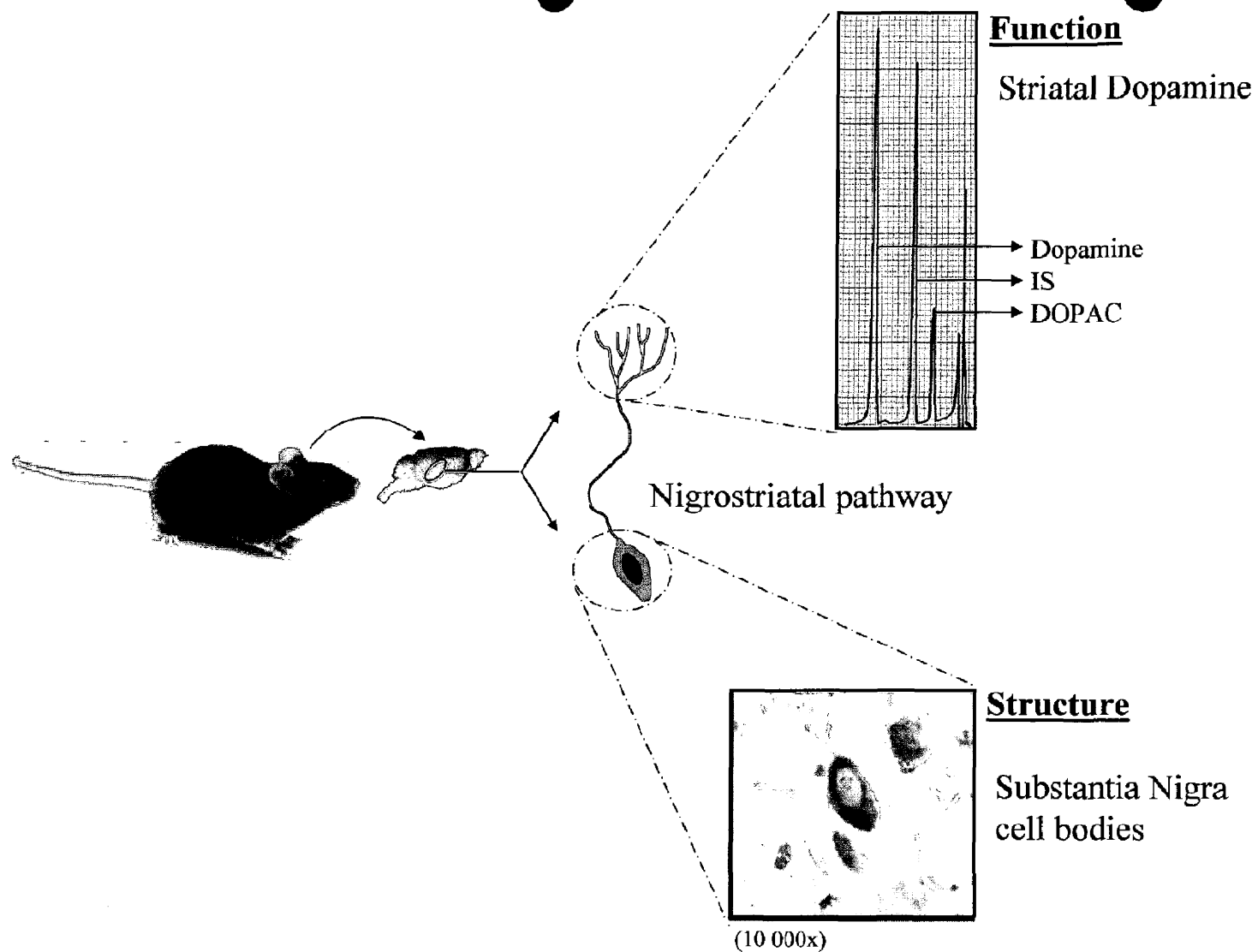
To assay for the functional inhibition of MAO-B *in vivo* we have examined the effects of TMN on the MAO-B mediated bioactivation of the parkinsonian inducing neurotoxin MPTP (protection against MPTP-induced depletion of striatal dopamine in C57BL/6 mice)

## METABOLIC BIOACTIVATION OF MPTP BY MAO-B

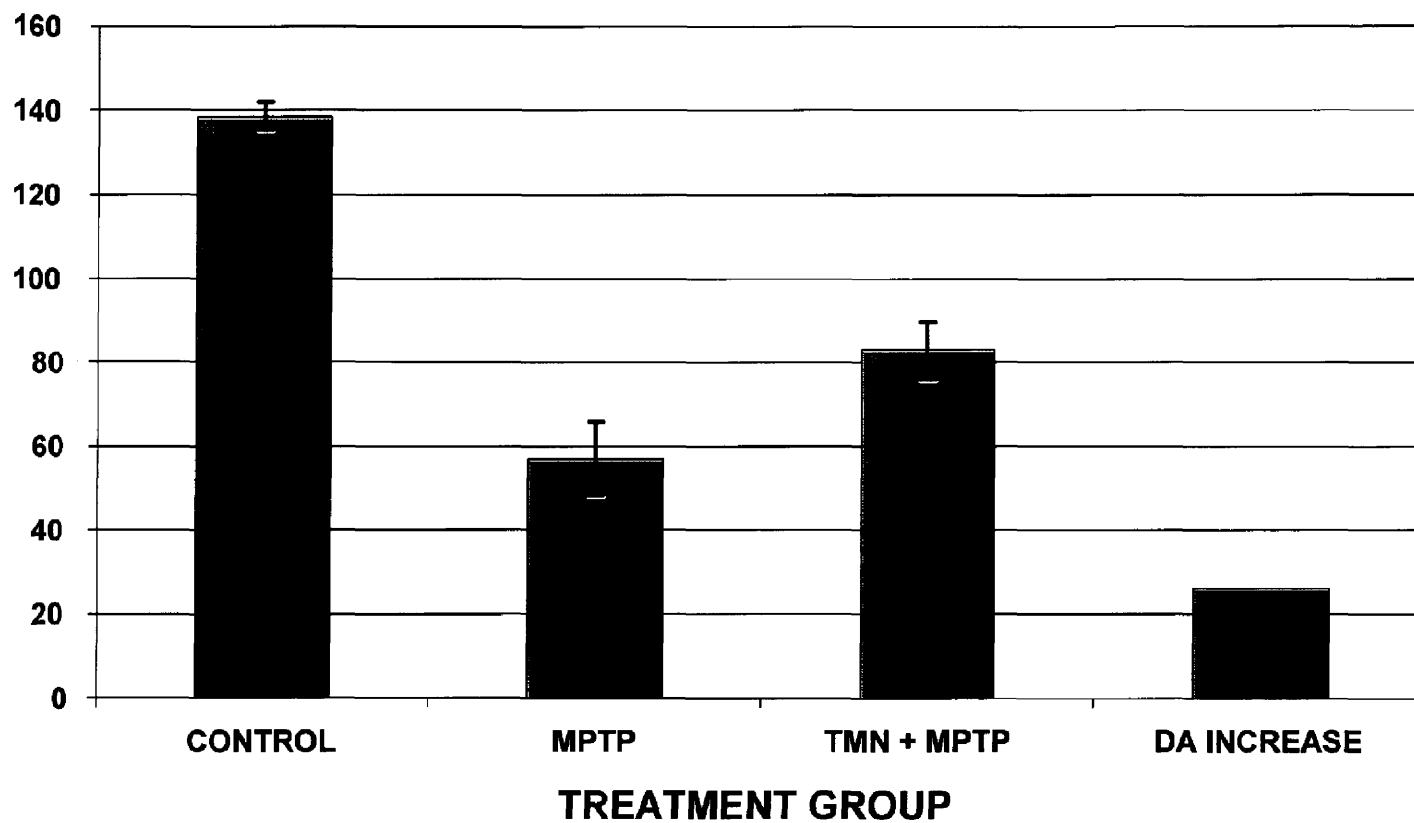


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**Figure 2. Relationship Between Striatal Dopamine and Substantia Nigra Cell bodies**

**STRIATAL DOPAMINE (pmol/mg)**

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## CONCLUSIONS

- (1) TOBACCO SMOKE CONSTITUENTS INHIBIT BOTH MAO-A AND MAO-B *IN VITRO*
- (2) THIS INHIBITION IS IN PART IRREVERSIBLE
- (3) EXPOSURE OF BOTH RATS AND MICE TO TOBACCO SMOKE LEADS TO THE (IRREVERSIBLE) INHIBITION OF BRAIN, KIDNEY, HEART AND LUNG MAO
- (4) CHROMATOGRAPHIC STUDIES INDICATE THE PRESENCE OF A VARIETY OF FRACTIONS WITH MAO INHIBITOR PROPERTIES
- (5) TMN (A MIXED A/B INHIBITOR) AND FARNESOL (A SELECTIVE MAO-B INHIBITOR) HAVE BEEN IDENTIFIED SO FAR
- (6) MUCH WORK REMAINS TO BE DONE

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## **ACKNOWLEDGMENTS**

### **ENZYME STUDIES**

Lindsey Aschenbach

Thangaraju Murugesan

### **RESEARCH CENTERS**

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BATELLE      Washington

PM FACILITY      Belgium

### **PHILIP MORRIS for FUNDING**

**BRUCE DAVIES**

**HARVEY W. PETERS CENTER**

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# Borgwaldt Automated Smoking Machine

QuickTime™ and a TIFF (Uncompressed) decompressor are needed to see this picture.

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**Objective:** To determine the effects of cigarette smoke exposure on brain MAO activity in an animal model.

**Rodent Model:** Rat {Sprague Dawley}

**Nose cone exposure of mainstream smoke**

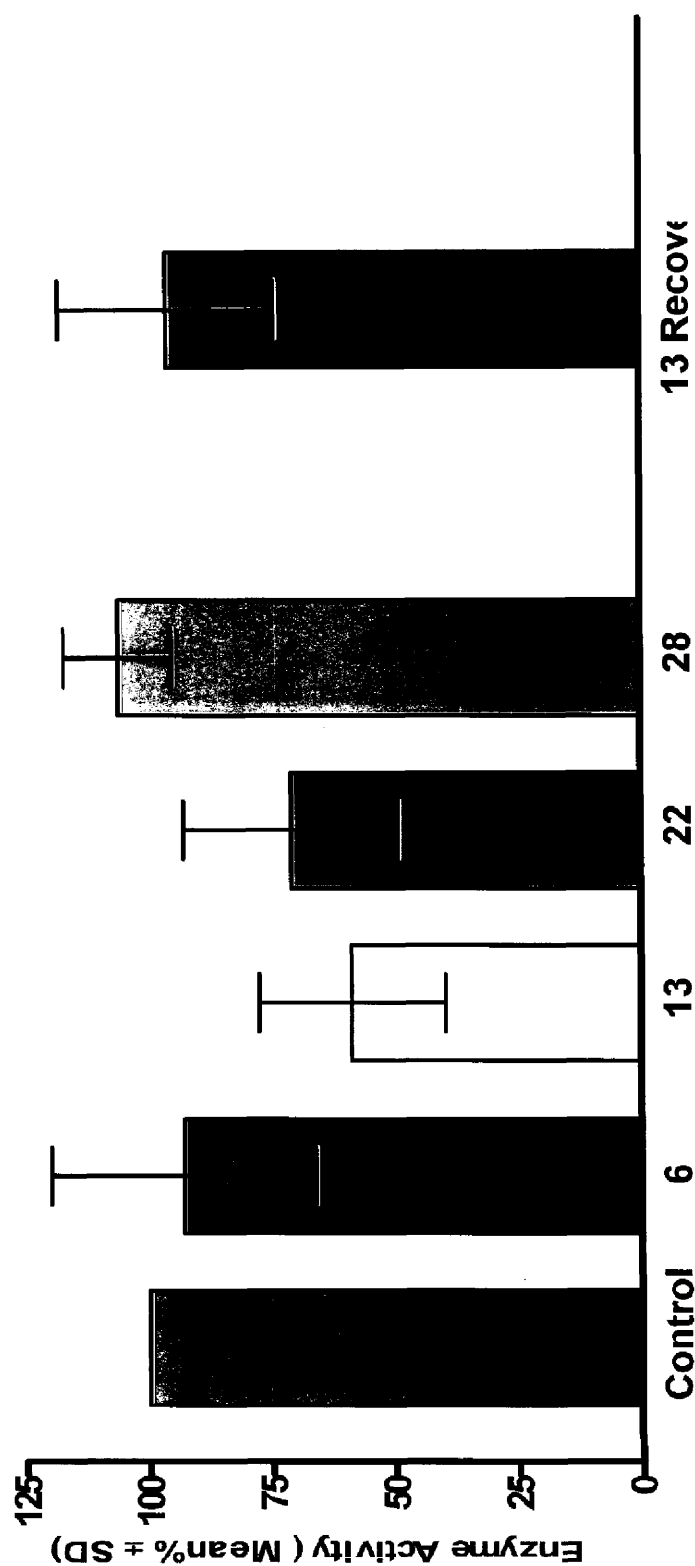
**Design:**

**Controls:** No exposure; Normal routine.

**Cage Controls:** Placed in smoke exposure chambers; exposed to normal air.

**Treated:** Placed in smoke exposure chambers. Exposed to smoke over specific time periods.

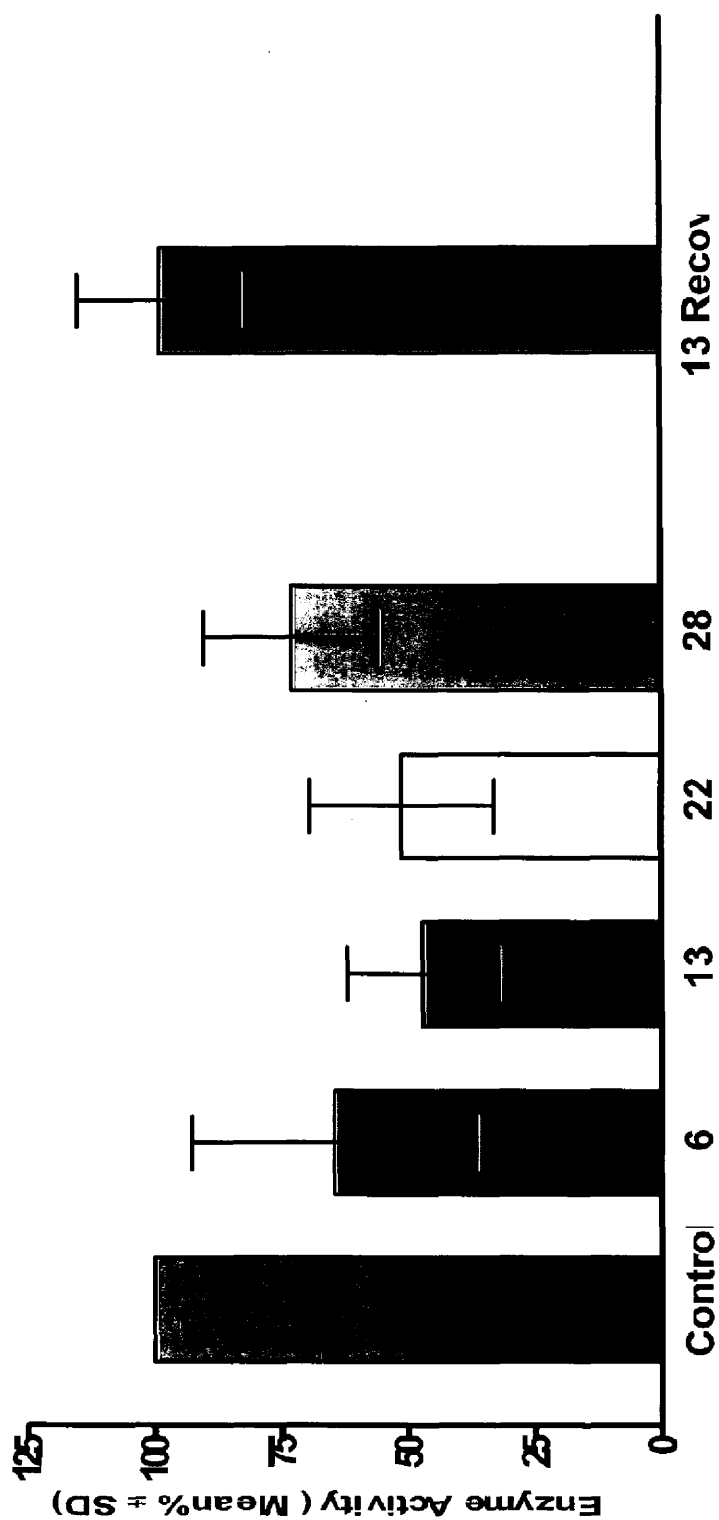
# ICR Mouse Brain MAO-A Activity at 600 µg/L Smoke



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# ICR Mouse Brain MAO-B Activity at 600 µg/L Smoke



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**C57 MICE**

**WHOLE BODY EXPOSURE** - One Dose; One Time Point - 13 weeks

**RECOVERY** several time points from 1 week up to 20 weeks

**MULTIPLE TISSUES**

Brain\*

Heart

Kidney

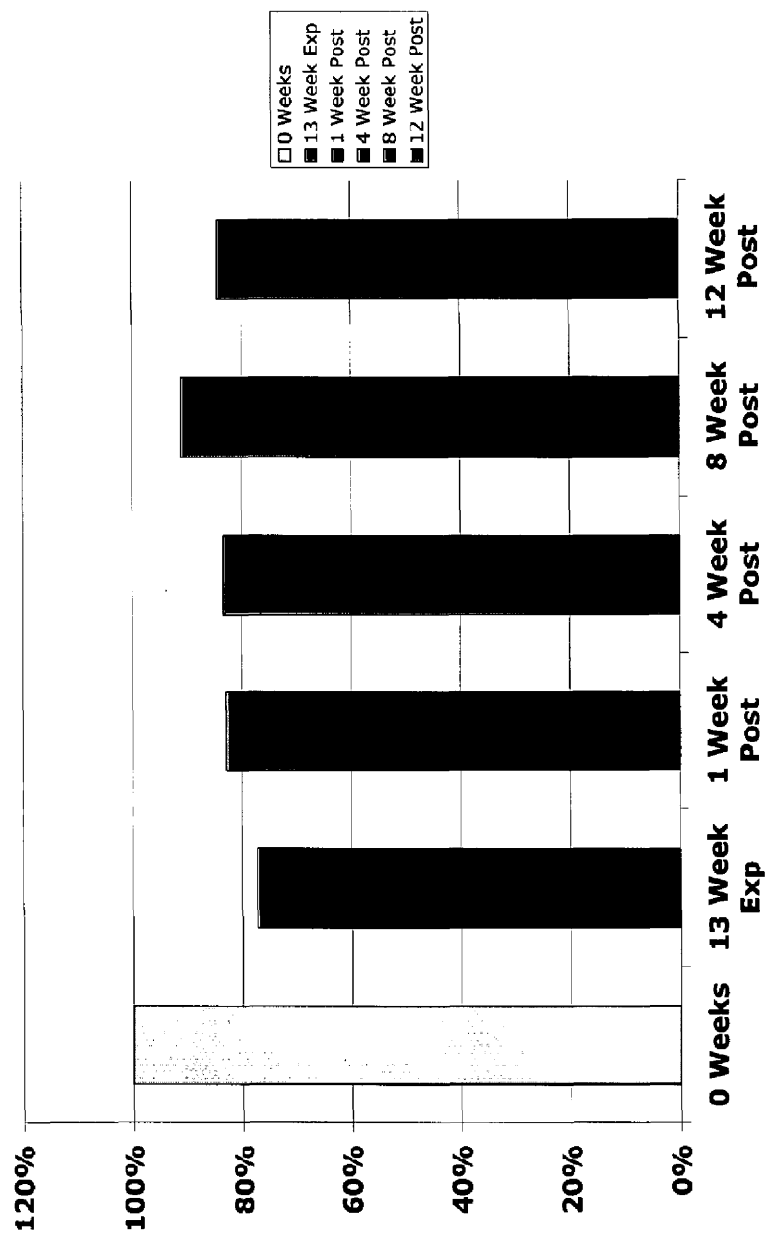
Liver\*

Lung

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## LUNG MAO-A VS. TIME



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